

5 more activity than the cognate, wild-type protein; and (d)
expressing the selected variant regulator protein in a
cell, thereby increasing production of the secondary
metabolite in the cell.

In certain embodiments of the fourth aspect, the cell
10 is a fungal cell. In certain embodiments of the fourth
aspect, the protein regulator of secondary metabolite
production is a transcription factor. In certain
embodiments of the fourth aspect, the protein regulator of
secondary metabolite production is a transmembrane
15 transporter, a protein that mediates secretion, a kinase,
a G-protein, a cell surface receptor, a GTPase activating
protein, a guanine nucleotide exchange factor, a
phosphatase, a protease, a phosphodiesterase, a bacterial
protein toxin, an importin, an RNA-binding protein, an SCF
20 complex component, an adherin, or a protein encoded within
a biosynthetic cluster. In certain embodiments of the
fourth aspect, the cell is a heterologous cell, preferably
selected from the group consisting of *S. cerevisiae*, *E.*
coli, *A. nidulans*, *Candida* sp., and *N. crassa*. In certain
25 other embodiments of the fourth aspect, the cell is a
homologous cell, preferably selected from the group
consisting of *Aspergillus* sp., *Penicillium* sp., *Acremonium*
chrysogenum, *Yarrowia lipolytica*, *Nodulisporium* sp.,
Fusarium sp., *Monascus* sp., *Claviceps* sp., *Trichoderma*
30 sp., *Tolypocladium* sp., *Tricotheicum* sp., *Fusidium* sp.,
Emericellopsis sp., *Cephalosporium* sp., *Cochliobolus* sp.,
Helminthosporium sp., *Agaricus brunescens*, *Ustilago*
maydis, *Neurospora* sp., *Pestalotiopsis* sp., and *Phaffia*
rhodozyma.

35 In certain other embodiments of the fourth aspect,
the cell is a heterologous cell and the method further
comprises expressing the variant regulator protein in a
homologous cell, thereby increasing secondary metabolite
production in the homologous cell. In certain embodiments
40 thereof, the heterologous cell is an organism selected
from the group consisting of *S. cerevisiae*, *E. coli*, *A.*

5 *nidulans*, *Candida* sp., , and *N. crassa* and the homologous
cell is an organism selected from the group consisting of
Aspergillus sp., *Penicillium* sp., *Acremonium chrysogenum*,
Yarrowia lipolytica, *Nodulisporium* sp., *Fusarium* sp.,
Monascus sp., *Claviceps* sp., *Trichoderma* sp.,
10 *Tolypocladium* sp., *Tricotheicium* sp., *Fusidium* sp.,
Emericellopsis sp., *Cephalosporium* sp., *Cochliobolus* sp.,
Helminthosporium sp., *Agaricus brunescens*, *Ustilago*
maydis, *Neurospora* sp., *Pestalotiopsis* sp. and *Phaffia*
rhodozyma.

15 In a fifth aspect, the invention provides an isolated
variant regulator protein of secondary metabolite
production having increased activity compared to a
cognate, wild-type protein, made by the process
comprising: (a) selecting a nucleic acid comprising a
20 polynucleotide encoding a protein regulator of secondary
metabolite production; (b) mutating the nucleic acid to
create a plurality of nucleic acid molecules encoding
variant regulator proteins of secondary metabolite
production; (c) selecting a variant regulator protein with
25 more activity than the cognate, wild-type protein; and (d)
recovering the selected variant regulator protein.

 In certain embodiments of the fifth aspect, the
variant regulator protein selected has more activity in a
fungal cell. In certain embodiments of the fifth aspect,
30 the protein regulator of secondary metabolite production
is a transcription factor. In certain embodiments of the
fifth aspect, the protein regulator of secondary
metabolite production is a transmembrane transporter, a
protein that mediates secretion, a kinase, a G-protein, a
35 cell surface receptor, a GTPase activating protein, a
guanine nucleotide exchange factor, a phosphatase, a
protease, a phosphodiesterase, a bacterial protein toxin,
an importin, an RNA-binding protein, an SCF complex
component, an adherin, or a protein encoded within a
40 biosynthetic cluster. In certain embodiments of the
fifth aspect, the variant regulator protein selected has

5 more activity in a heterologous cell, preferably selected from the group consisting of *S. cerevisiae*, *E. coli*, *A. nidulans*, *Candida* sp., *Neurospora* sp., *Pestalotiopsis* sp., and *N. crassa*. In certain embodiments of the fifth aspect, the variant regulator protein selected has more
10 activity in a homologous cell, preferably selected from the group consisting of *Aspergillus* sp., *Penicillium* sp., *Acremonium chrysogenum*, *Yarrowia lipolytica*, *Nodulisporium* sp., *Fusarium* sp., *Monascus* sp., *Claviceps* sp., *Trichoderma* sp., *Tolypocladium* sp., *Tricotheicum* sp.,
15 *Fusidium* sp., *Emericellopsis* sp., *Cephalosporium* sp., *Cochliobolus* sp., *Helminthosporium* sp., *Agaricus brunescens*, *Ustilago maydis*, *Neurospora* sp., *Pestalotiopsis* sp., and *Phaffia rhodozyma*.

In certain embodiments of the fifth aspect, the
20 variant regulator protein selected has more activity in a homologous cell and a heterologous cell. In embodiments thereof, the heterologous cell is an organism selected from the group consisting of *S. cerevisiae*, *E. coli*, *A. nidulans*, *Candida* sp., *Neurospora* sp., *Pestalotiopsis* sp., and *N. crassa* and the homologous cell is an organism selected from the group consisting of *Aspergillus* sp., *Penicillium* sp., *Acremonium chrysogenum*, *Yarrowia lipolytica*, *Nodulisporium* sp., *Fusarium* sp., *Monascus* sp., *Claviceps* sp., *Trichoderma* sp., *Tolypocladium* sp.,
25 *Tricotheicum* sp., *Fusidium* sp., *Emericellopsis* sp., *Cephalosporium* sp., *Cochliobolus* sp., *Helminthosporium* sp., *Agaricus brunescens*, *Ustilago maydis*, *Neurospora* sp., *Pestalotiopsis* sp., and *Phaffia rhodozyma*.

In yet another embodiment of the fifth aspect, the
35 variant regulator protein is a variant protein of the lovE protein having at least one of the following mutations:
(1) a Group 6 amino acid residue mutated to a Group 2 amino acid residue at position 31, for example, the mutation represented by F31L; (2) a Group 3 amino acid residue mutated to a Group 5 amino acid residue at position 41, for example, the mutation represented by Q41K